

10/019284

1 531 Rec'd PCT. 02 JAN 2002

## SEQUENCE LISTING

&lt;110&gt; Ajinomoto Co., Inc.

&lt;120&gt; DNA Encoding Sucrose PTS Enzyme II

&lt;130&gt; B644MSOP1027

&lt;150&gt; JP 11-189512

&lt;151&gt; 1999-07-02

&lt;160&gt; 21

&lt;170&gt; PatentIn Ver. 2.0

&lt;210&gt; 1

&lt;211&gt; 5969

&lt;212&gt; DNA

&lt;213&gt; Brevibacterium lactofermentum

&lt;220&gt;

&lt;221&gt; CDS

&lt;222&gt; (3779).. (5761)

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 Met Asp His Lys Asp Leu Ala Gln Arg Ile Leu Arg Asp Ile Gly Gly  
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 gaa gac aac att gtc gcc gcc gca cac tgt gca acg cgt tta cgc ctc 3874  
 Glu Asp Asn Ile Val Ala Ala Ala His Cys Ala Thr Arg Leu Arg Leu  
                     20                    25                    30  
 gtg ctc aaa gac acc aag gat gtg gat cgc caa agt ctg gat gat gat 3922  
 Val Leu Lys Asp Thr Lys Asp Val Asp Arg Gln Ser Leu Asp Asp Asp  
                     35                    40                    45  
 cca gat ctg aaa ggc acc ttt gaa act ggc ggc atg ttc cag atc atc 3970  
 Pro Asp Leu Lys Gly Thr Phe Glu Thr Gly Gly Met Phe Gln Ile Ile  
                     50                    55                    60  
 gtc ggg cca ggc gat gtg gat cat gtt ttc aaa gaa ctc gat gac gca 4018  
 Val Gly Pro Gly Asp Val Asp His Val Phe Lys Glu Leu Asp Asp Ala  
                     65                    70                    75                    80  
 acc tcc aaa gac atc gct gtg tcc aca gag cag ctc aaa gat gtt gtg 4066  
 Thr Ser Lys Asp Ile Ala Val Ser Thr Glu Gln Leu Lys Asp Val Val  
                     85                    90                    95  
 gct aac aac gcc aac tgg ttc agc cgt gct gtg aag gta ttg gcg gac 4114  
 Ala Asn Asn Ala Asn Trp Phe Ser Arg Ala Val Lys Val Leu Ala Asp  
                     100                    105                    110  
 att ttc gtc ccg ctg att cca atc ttg gtt ggt ggc ggt ctg ctc atg 4162  
 Ile Phe Val Pro Leu Ile Pro Ile Leu Val Gly Gly Gly Leu Leu Met  
                     115                    120                    125  
 gct atc aac aat gtg ttg gtt gcg cag gat ctg ttc ggt ccg caa tca 4210  
 Ala Ile Asn Asn Val Leu Val Ala Gln Asp Leu Phe Gly Pro Gln Ser  
                     130                    135                    140  
 ctg gtg gag atg ttc cct cag atc agc ggt gtt gct gag atg atc aac 4258  
 Leu Val Glu Met Phe Pro Gln Ile Ser Gly Val Ala Glu Met Ile Asn  
                     145                    150                    155                    160  
 ctg atg gca tct gcg ccg ttc gcg ttc ttg cca gtg ttg gtt ggt ttc 4306  
 Leu Met Ala Ser Ala Pro Phe Ala Phe Leu Pro Val Leu Val Gly Phe  
                     165                    170                    175

acc gca acc aag cgt ttc ggt ggc aat gag ttc ctg ggc gcc ggc att 4354  
 Thr Ala Thr Lys Arg Phe Gly Gly Asn Glu Phe Leu Gly Ala Gly Ile  
 180 185 190  
 ggt atg gcg atg gtg ttc cca acc ctg gtt aac ggc tac gac gtg gcc 4402  
 Gly Met Ala Met Val Phe Pro Thr Leu Val Asn Gly Tyr Asp Val Ala  
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 gcc acc atg acc gcg ggc gaa atg cca atg tgg tcc ctg ttt ggt ttg 4450  
 Ala Thr Met Thr Ala Gly Glu Met Pro Met Trp Ser Leu Phe Gly Leu  
 210 215 220  
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 Asp Val Ala Gln Ala Gly Tyr Gln Gly Thr Val Leu Pro Val Leu Val  
 225 230 235 240  
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 Val Ser Trp Ile Leu Ala Thr Ile Glu Lys Phe Leu His Lys Arg Leu  
 245 250 255  
 atg ggc act gca gac ttc ctg atc acc cca gtg ttg act ctg ctg ctc 4594  
 Met Gly Thr Ala Asp Phe Leu Ile Thr Pro Val Leu Thr Leu Leu Leu  
 260 265 270  
 acc ggc ttc ctt acg ttc att gct att ggt cca gca atg cgc tgg gtg 4642  
 Thr Gly Phe Leu Thr Phe Ile Ala Ile Gly Pro Ala Met Arg Trp Val  
 275 280 285  
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 Gly Asp Leu Leu Ala His Gly Leu Gln Gly Leu Tyr Asp Phe Gly Gly  
 290 295 300  
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 Pro Val Gly Gly Leu Leu Phe Gly Leu Val Tyr Ser Pro Ile Val Ile  
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 Thr Gly Leu His Gln Ser Phe Pro Pro Ile Glu Leu Glu Leu Phe Asn  
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 Gln Gly Gly Ser Phe Ile Phe Ala Thr Ala Ser Met Ala Asn Ile Ala  
 340 345 350  
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 Gln Gly Ala Ala Cys Leu Ala Val Phe Phe Leu Ala Lys Ser Glu Lys  
 355 360 365  
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 Leu Lys Gly Leu Ala Gly Ala Ser Gly Val Ser Ala Val Leu Gly Ile  
 370 375 380  
 aca gag cct gcg atc ttc ggt gtg aac ctt cgc ctg cgc tgg ccg ttc 4978  
 Thr Glu Pro Ala Ile Phe Gly Val Asn Leu Arg Leu Arg Trp Pro Phe

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tac att ggt atc ggt acc gca gct atc ggt ggc gct ttg att gca ctc				5026
Tyr Ile Gly Ile Gly Thr Ala Ala Ile Gly Gly Ala Leu Ile Ala Leu				
	405	410	415	
ttt gat atc aag gca gtt gcg ttg ggc gct gca ggt ttc ttg ggt gtt				5074
Phe Asp Ile Lys Ala Val Ala Leu Gly Ala Ala Gly Phe Leu Gly Val				
	420	425	430	
gtt tct att gat gct cca gat atg gtc atg ttc ttg gtt tgc gcg gta				5122
Val Ser Ile Asp Ala Pro Asp Met Val Met Phe Leu Val Cys Ala Val				
	435	440	445	
gtt acc ttt gtc atc gca ttc ggc gca gcg att gct tat ggc ctt tac				5170
Val Thr Phe Val Ile Ala Phe Gly Ala Ala Ile Ala Tyr Gly Leu Tyr				
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Leu Val Arg Arg Asn Gly Ser Ile Asp Pro Asp Ala Thr Ala Ala Pro				
465	470	475	480	
gtg cct gca gga acg acc aaa gcc gaa gca gaa gca ccc gca gaa ttt				5266
Val Pro Ala Gly Thr Thr Lys Ala Glu Ala Glu Ala Pro Ala Glu Phe				
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tca aac gat tcc acc atc atc cag gca cct ttg acc ggt gaa gct atc				5314
Ser Asn Asp Ser Thr Ile Ile Gln Ala Pro Leu Thr Gly Glu Ala Ile				
	500	505	510	
gca ctg agc agc gtc agc gat gcc atg ttt gcc agc gga aag ctt ggc				5362
Ala Leu Ser Ser Val Ser Asp Ala Met Phe Ala Ser Gly Lys Leu Gly				
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tca ggt gtt gcg atc gtc ccc acc aag ggg cag ctg gtt tca cca gtg				5410
Ser Gly Val Ala Ile Val Pro Thr Lys Gly Gln Leu Val Ser Pro Val				
	530	535	540	
agc gga aag atc gtg gtg gcc ttc cca tct ggt cac gct ttc gca gtc				5458
Ser Gly Lys Ile Val Val Ala Phe Pro Ser Gly His Ala Phe Ala Val				
545	550	555	560	
cgc act aag gct gag gat ggt tcc aat gtg gat atc ttg atg cac att				5506
Arg Thr Lys Ala Glu Asp Gly Ser Asn Val Asp Ile Leu Met His Ile				
	565	570	575	
ggt ttc gac acc gta aac ctc aac ggc acg cac ttt aac ccg ctg aag				5554
Gly Phe Asp Thr Val Asn Leu Asn Gly Thr His Phe Asn Pro Leu Lys				
	580	585	590	
aag cag ggc gat gaa gtc aaa gca ggg gag ctg ctg tgt gaa ttc gat				5602
Lys Gln Gly Asp Glu Val Lys Ala Gly Glu Leu Leu Cys Glu Phe Asp				
	595	600	605	
att gat gcc att aag gct gca ggt tat gag gta acc acg ccg att gtt				5650

Ile Asp Ala Ile Lys Ala Ala Gly Tyr Glu Val Thr Thr Pro Ile Val  
 610 615 620  
 gtt tcg aat tac aag aaa acc gga cct gta aac act tac ggt ttg ggc 5698  
 Val Ser Asn Tyr Lys Lys Thr Gly Pro Val Asn Thr Tyr Gly Leu Gly  
 625 630 635 640  
 gaa att gaa gcg gga gcc aac ctg ctc aac gtc gca aag aaa gaa gcg 5746  
 Glu Ile Glu Ala Gly Ala Asn Leu Leu Asn Val Ala Lys Lys Glu Ala  
 645 650 655  
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 Val Pro Ala Thr Pro  
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&lt;210&gt; 2

&lt;211&gt; 661

&lt;212&gt; PRT

&lt;213&gt; Brevibacterium lactofermentum

&lt;400&gt; 2

Met Asp His Lys Asp Leu Ala Gln Arg Ile Leu Arg Asp Ile Gly Gly  
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 Glu Asp Asn Ile Val Ala Ala Ala His Cys Ala Thr Arg Leu Arg Leu  
 20 25 30  
 Val Leu Lys Asp Thr Lys Asp Val Asp Arg Gln Ser Leu Asp Asp Asp  
 35 40 45  
 Pro Asp Leu Lys Gly Thr Phe Glu Thr Gly Gly Met Phe Gln Ile Ile  
 50 55 60  
 Val Gly Pro Gly Asp Val Asp His Val Phe Lys Glu Leu Asp Asp Ala  
 65 70 75 80  
 Thr Ser Lys Asp Ile Ala Val Ser Thr Glu Gln Leu Lys Asp Val Val  
 85 90 95  
 Ala Asn Asn Ala Asn Trp Phe Ser Arg Ala Val Lys Val Leu Ala Asp  
 100 105 110  
 Ile Phe Val Pro Leu Ile Pro Ile Leu Val Gly Gly Gly Leu Leu Met  
 115 120 125  
 Ala Ile Asn Asn Val Leu Val Ala Gln Asp Leu Phe Gly Pro Gln Ser  
 130 135 140  
 Leu Val Glu Met Phe Pro Gln Ile Ser Gly Val Ala Glu Met Ile Asn  
 145 150 155 160

Leu Met Ala Ser Ala Pro Phe Ala Phe Leu Pro Val Leu Val Gly Phe  
 165 170 175  
 Thr Ala Thr Lys Arg Phe Gly Gly Asn Glu Phe Leu Gly Ala Gly Ile  
 180 185 190  
 Gly Met Ala Met Val Phe Pro Thr Leu Val Asn Gly Tyr Asp Val Ala  
 195 200 205  
 Ala Thr Met Thr Ala Gly Glu Met Pro Met Trp Ser Leu Phe Gly Leu  
 210 215 220  
 Asp Val Ala Gln Ala Gly Tyr Gln Gly Thr Val Leu Pro Val Leu Val  
 225 230 235 240  
 Val Ser Trp Ile Leu Ala Thr Ile Glu Lys Phe Leu His Lys Arg Leu  
 245 250 255  
 Met Gly Thr Ala Asp Phe Leu Ile Thr Pro Val Leu Thr Leu Leu Leu  
 260 265 270  
 Thr Gly Phe Leu Thr Phe Ile Ala Ile Gly Pro Ala Met Arg Trp Val  
 275 280 285  
 Gly Asp Leu Leu Ala His Gly Leu Gln Gly Leu Tyr Asp Phe Gly Gly  
 290 295 300  
 Pro Val Gly Gly Leu Leu Phe Gly Leu Val Tyr Ser Pro Ile Val Ile  
 305 310 315 320  
 Thr Gly Leu His Gln Ser Phe Pro Pro Ile Glu Leu Glu Leu Phe Asn  
 325 330 335  
 Gln Gly Gly Ser Phe Ile Phe Ala Thr Ala Ser Met Ala Asn Ile Ala  
 340 345 350  
 Gln Gly Ala Ala Cys Leu Ala Val Phe Phe Leu Ala Lys Ser Glu Lys  
 355 360 365  
 Leu Lys Gly Leu Ala Gly Ala Ser Gly Val Ser Ala Val Leu Gly Ile  
 370 375 380  
 Thr Glu Pro Ala Ile Phe Gly Val Asn Leu Arg Leu Arg Trp Pro Phe  
 385 390 395 400  
 Tyr Ile Gly Ile Gly Thr Ala Ala Ile Gly Gly Ala Leu Ile Ala Leu  
 405 410 415  
 Phe Asp Ile Lys Ala Val Ala Leu Gly Ala Ala Gly Phe Leu Gly Val  
 420 425 430  
 Val Ser Ile Asp Ala Pro Asp Met Val Met Phe Leu Val Cys Ala Val  
 435 440 445  
 Val Thr Phe Val Ile Ala Phe Gly Ala Ala Ile Ala Tyr Gly Leu Tyr  
 450 455 460  
 Leu Val Arg Arg Asn Gly Ser Ile Asp Pro Asp Ala Thr Ala Ala Pro  
 465 470 475 480  
 Val Pro Ala Gly Thr Thr Lys Ala Glu Ala Glu Ala Pro Ala Glu Phe

	485		490		495
Ser Asn Asp	Ser Thr Ile Ile Gln Ala	Pro Leu Thr Gly Glu Ala Ile			
	500		505		510
Ala Leu Ser	Ser Val Ser Asp Ala Met Phe Ala Ser	Gly Lys Leu Gly			
	515		520		525
Ser Gly Val	Ala Ile Val Pro Thr Lys Gly Gln Leu Val	Ser Pro Val			
	530		535		540
Ser Gly Lys	Ile Val Val Ala Phe Pro Ser Gly His Ala Phe Ala Val				
545		550		555	560
Arg Thr Lys	Ala Glu Asp Gly Ser Asn Val Asp Ile Leu Met His Ile				
	565		570		575
Gly Phe Asp	Thr Val Asn Leu Asn Gly Thr His Phe Asn Pro Leu Lys				
	580		585		590
Lys Gln Gly	Asp Glu Val Lys Ala Gly Glu Leu Leu Cys Glu Phe Asp				
	595		600		605
Ile Asp Ala	Ile Lys Ala Ala Gly Tyr Glu Val Thr Thr Pro Ile Val				
	610		615		620
Val Ser Asn	Tyr Lys Lys Thr Gly Pro Val Asn Thr Tyr Gly Leu Gly				
625		630		635	640
Glu Ile Glu	Ala Gly Ala Asn Leu Leu Asn Val Ala Lys Lys Glu Ala				
	645		650		655
Val Pro Ala	Thr Pro				
	660				

&lt;210&gt; 3

&lt;211&gt; 44

&lt;212&gt; DNA

&lt;213&gt; Artificial Sequence

&lt;220&gt;

&lt;223&gt; Description of Artificial Sequence: Sau3AI cassette

&lt;220&gt;

&lt;221&gt; misc\_feature

&lt;222&gt; (44)

<223> complementary strand extends a single strand having  
a sequence of 3'-ctag-5' at this position in the  
direction of 5' from 3'

&lt;400&gt; 3

gtacatattg tcgttagaac gcgtaatacg actcactata ggga



<210> 4  
<211> 47  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: EcoRI cassette

<220>  
<221> misc\_feature  
<222> (47)  
<223> complementary strand extends a single strand having  
a sequence of 3'-ttaa-5' at this position in the  
direction of 5' from 3'

<400> 4  
gtacatatgt tcgttagaac gcgtaatacg actcactata gggagag 47

<210> 5  
<211> 46  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: HindIII cassette

<220>  
<221> misc\_feature  
<222> (46)  
<223> complementary strand extends a single strand having  
a sequence of 3'-tcga-5' at this position in the  
direction of 5' from 3'

<400> 5  
gtacatatgt tcgttagaac gcgtaatacg actcactata gggaga 46

<210> 6  
<211> 51  
<212> DNA  
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PstI cassette

<220>

<221> misc\_feature

<222> (48)..(51)

<223> complementary strand does not exist

<400> 6

gtacatatg tcgttagaac gcgtaatacg actcactata gggagactgc a

51

<210> 7

<211> 47

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: SalI cassette

<220>

<221> misc\_feature

<222> (47)

<223> complementary strand extends a single strand having  
a sequence of 3'-agct-5' at this position in the  
direction of 5' from 3'

<400> 7

gtacatatg tcgttagaac gcgtaatacg actcactata gggagag

47

<210> 8

<211> 47

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: XbaI cassette

<220>

<221> misc\_feature

<222> (47)

<223> complementary strand extends a single strand having

a sequence of 3'-gatc-5' at this position in the  
direction of 5' from 3'

<400> 8  
gtacatatg tcgttagaac gcgtaatacg actcactata gggagat 47

<210> 9  
<211> 25  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: primer for PCR

<400> 9  
cgtcttgcga ggattcagcg agctg 25

<210> 10  
<211> 25  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: primer for PCR

<400> 10  
agctggattt cggccatgaa ttcta 25

<210> 11  
<211> 23  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: primer for PCR

<400> 11  
gatctgttcg gtccgcaatc act 23

<210> 12  
<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer for PCR

<400> 12

cactggtgga gatgttcct cagat

25

<210> 13

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer for PCR

<400> 13

catcttcgca accgcatcca tggcc

25

<210> 14

<211> 24

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer for PCR

<400> 14

cgcgagggt gcagcatggt tggc

24

<210> 15

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer for PCR

<400> 15

gggccttgca ggtgcttcag gtgtc

25

<210> 16  
<211> 25  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: primer for PCR

<400> 16  
ccgctgttct tggattaca gagcc 25

<210> 17  
<211> 25  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: primer for PCR

<400> 17  
gcagcgtcag cgatgccatg ttgc 25

<210> 18  
<211> 25  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: primer for PCR

<400> 18  
gcttggtca ggtgttcga tcgtc 25

<210> 19  
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<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence: cassette  
primer 1

<400> 19

gtacatatgt tcgttagaac gcggtaatat gactca

36

<210> 20

<211> 35

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: cassette  
primer 2

<400> 20

cgttagaacg cgtaatacga ctactatag ggaga

35

<210> 21

<211> 24

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer for PCR

<400> 21

cgctactgct gaacgaacat gtcc

24